

Appl. No. 09/924,197

Amdt dated July 21, 2005

Reply to Office action of March 23, 2005

Amendments to the Claims

This listing of claims will replace all prior versions and listings of claims in this application:

1. (currently amended) A method of reducing expression of a target gene in a plant cell, the method comprising the step of expressing in the cell an expression cassette comprising a promoter operably linked to a sense or antisense targeting sequence having at least about 80% identity to at least a subsequence of the target gene, wherein the subsequence has a length of at least about 25 nucleotides, and an inverted repeat prepared from ~~[[of]]~~ a subsequence ~~of an~~ of a nopaline synthase (NOS) gene ~~[[.]]~~;

wherein the inverted repeat comprises:

a sense element comprising the subsequence from the NOS gene in a sense orientation; and

a antisense element comprising the subsequence from the NOS gene in an antisense orientation;

and

the inverted repeat is at least about 30 base pairs in length and ~~[[is]]~~ heterologous to the targeting sequence ~~[[.]]~~;

thereby reducing expression of the target gene.

2. (previously presented) The method of claim 1, wherein the inverted repeat is in a position 3' to the targeting sequence.

3. (previously presented) The method of claim 1, wherein the inverted repeat is in a position 5' to the targeting sequence.

4. (previously presented) The method of claim 1, wherein the inverted repeat is from the 3' untranslated region of the NOS gene.

5. (previously presented) The method of claim 4, wherein the inverted repeat is from the terminator region of the NOS gene.

6. (previously presented) The method of claim 1, wherein the inverted repeat is from the 5' untranslated region of the NOS gene.

7. (previously presented) The method of claim 1, wherein the inverted repeat is from the coding region of the NOS gene.

Appl. No. 09/924,197
Amdt dated July 21, 2005
Reply to Office action of March 23, 2005

8. (currently amended) The method of claim 1, wherein the NOS gene is from an *Agrobacterium* [[sp.]] species.
9. (previously presented) The method of claim 1, wherein the inverted repeat comprises a sense region, a linker region, and an antisense region.
10. (previously presented) The method of claim 1, wherein the inverted repeat is from about 30 to about 200 nucleotides in length.
11. (previously presented) The method of claim 1, wherein the targeting sequence is a sense sequence.
12. (previously presented) The method of claim 1, wherein the targeting sequence is an antisense sequence.
13. (previously presented) The method of claim 1, wherein the targeting sequence has substantial identity to a plant pathogen target gene.
14. (previously presented) The method of claim 13, wherein the targeting sequence is a viral sequence, a bacterial sequence, an insect sequence, a fungal sequence, or a nematode sequence.
15. (currently amended) The method of claim 1, wherein the targeting sequence has substantial at least about 85% identity to a plant target gene.
16. (previously presented) The method of claim 1, wherein the targeting sequence is from about 100 to about 1000 nucleotides in length.
17. (previously presented) The method of claim 1 wherein the targeting sequence is from a coding region of the target gene.
18. (previously presented) The method of claim 1, wherein the targeting sequence is from a 5' untranslated region of the target gene.
19. (previously presented) The method of claim 1, wherein the targeting sequence is from a 3' untranslated region of the target gene.

Appl. No. 09/924,197

Amdt dated July 21, 2005

Reply to Office action of March 23, 2005

20. (previously presented) The method of claim 1, wherein the target gene is polygalacturonase.
21. (previously presented) The method of claim 1, wherein the promoter is a tissue specific promoter.
22. (previously presented) The method of claim 1, wherein the promoter is a plant promoter.
23. (previously presented) The method of claim 22, wherein the promoter is a cauliflower mosaic virus 35S promoter or a figwort mosaic virus 34S promoter.
24. (canceled)
25. (previously presented) The method of claim 1, wherein the plant is selected from the group consisting of wheat, corn, rice, sorghum, pepper, tomato, squash, banana, strawberry, carrot, bean, cabbage, beet, cotton, grape, pea, pineapple, potato, soybean, yam, and alfalfa.
26. (previously presented) The method of claim 1, wherein the expression cassette has a nucleotide sequence of SEQ ID NO: 1.
27. (previously presented) The method of claim 1, wherein the targeting sequence comprises a premature stop codon that inhibits translation of the targeting sequence.
28. (withdrawn) An expression cassette comprising a promoter operably linked to a sense or antisense targeting sequence having substantial identity to at least a subsequence of the target gene, and an inverted repeat of a subsequence of an NOS gene, wherein the inverted repeat is heterologous to the targeting sequence.
29. (withdrawn) The expression cassette of claim 28, wherein the inverted repeat is in a position 3' to the targeting sequence.
30. (withdrawn) The expression cassette of claim 28, wherein the inverted repeat is in a position 5' to the targeting sequence.

Appl. No. 09/924,197
Amdt dated July 21, 2005
Reply to Office action of March 23, 2005

31. (withdrawn) The expression cassette of claim 28, wherein the inverted repeat is from the 3' untranslated region of the NOS gene.

32. (withdrawn) The expression cassette of claim 31, wherein the inverted repeat is from the terminator region of the NOS gene.

33. (withdrawn) The expression cassette of claim 28, wherein the inverted repeat is from the 5' untranslated region of the NOS gene.

34. (withdrawn) The expression cassette of claim 28, wherein the inverted repeat is from the coding region of the NOS gene.

35. (withdrawn) The expression cassette of claim 28, wherein the NOS gene is from an *Agrobacterium* sp,

36. (withdrawn) The expression cassette of claim 28, wherein the inverted repeat comprises a sense region, a linker region, and an antisense region.

37. (withdrawn) The expression cassette of claim 28, wherein the inverted repeat is from about 30 to about 200 nucleotides in length.

38. (withdrawn) The expression cassette of claim 28, wherein the targeting sequence is a sense sequence.

39. (withdrawn) The expression cassette of claim 28, wherein the targeting sequence is an antisense sequence.

40. (withdrawn) The expression cassette of claim 28, wherein the targeting sequence has substantial identity to a plant pathogen target gene.

41. (withdrawn) The expression cassette of claim 40, wherein the targeting sequence is a viral sequence, a bacterial sequence, an insect sequence, a fungal sequence, or a nematode sequence.

42. (withdrawn) The expression cassette of claim 28, wherein the targeting sequence has substantial identity to a plant target gene.

Appl. No. 09/924,197

Amdt dated July 21, 2005

Reply to Office action of March 23, 2005

43. (withdrawn) The expression cassette of claim 28, wherein the targeting sequence is from about 100 to about 1000 nucleotides in length.

44. (withdrawn) The expression cassette of claim 28, wherein the targeting sequence is from a coding region of the target gene.

45. (withdrawn) The expression cassette of claim 28, wherein the targeting sequence is from a 5' untranslated region of the target gene.

46. (withdrawn) The expression cassette of claim 28, wherein the targeting sequence is from a 3' untranslated region of the target gene.

47. (withdrawn) The expression cassette of claim 42, wherein the target gene is polygalacturonase.

48. (withdrawn) The expression cassette of claim 28, wherein the promoter is a tissue specific promoter.

49. (withdrawn) The expression cassette of claim 28, wherein the promoter is a plant promoter.

50. (withdrawn) The expression cassette of claim 49, wherein the promoter is a cauliflower mosaic virus 35S promoter or a figwort mosaic virus 34S promoter.

51. (withdrawn) The expression cassette of claim 28, wherein the expression cassette has a nucleotide sequence of SEQ ID NO: 1.

52. (withdrawn) The expression cassette of claim 28, wherein the targeting sequence comprises a premature stop codon that inhibits translation of the targeting sequence.

53. (withdrawn) A transgenic plant comprising the expression cassette of claim 28.